

What is claimed is:

1. An improved method for purification of a Factor VIII polypeptide comprising:
 - a) adding a mixture containing Factor VIII polypeptide to be purified to an
 - 5 immunoaffinity matrix which binds by hydrophobic attraction to the FVIII polypeptide;
 - b) eluting the Factor VIII polypeptide from the immunoaffinity matrix with a desorbing solution which causes desorption of the Factor VIII polypeptide, which is released in an elution solution;
 - c) diluting the desorbing solution with a solution comprising higher ionic strength
 - 10 than that of the elution solution, resulting in a diluted Factor VIII solution;
 - d) passing the diluted Factor VIII solution through an ion exchange column capable of binding to the Factor VIII polypeptide, thereby binding the Factor VIII polypeptide while allowing contaminants to pass through the ion exchange column; and
 - e) eluting the purified Factor VIII polypeptide from the ion exchange column.
- 15 2. A method of claim 1, wherein the desorbing solution of step (b) contains no salt, and the dilution of step (c) is performed using a solution comprising from about 7 to about 20 mM NaCl.
3. A method of claim 1, wherein the desorbing solution of step (b) contains no salt, and the dilution of step (c) is performed using a solution comprising about 15 mM NaCl.
- 20 4. A method of claim 3, wherein the desorbing solution is diluted from about 3-fold to about 5-fold.
5. A method of claim 3, wherein the desorbing solution is diluted about 3-fold.
6. An improved method for purification of a Factor VIII polypeptide comprising:
 - a) adding a mixture containing Factor VIII polypeptide to be purified to an
 - 25 immunoaffinity matrix which binds by hydrophobic attraction to the FVIII polypeptide;
 - b) eluting the Factor VIII polypeptide from the immunoaffinity matrix with a desorbing solution which causes desorption of the Factor VIII polypeptide, which is released in an elution solution, wherein the desorbing solution comprises a non-polar agent;
 - c) diluting the elution solution with a solution comprising lower concentration of the
 - 30 non-polar agent than that of the elution solution, resulting in a diluted Factor VIII solution;

d) passing the diluted Factor VIII solution through an ion exchange column capable of binding to the Factor VIII polypeptide, thereby binding the Factor VIII polypeptide while allowing contaminants to pass through the ion exchange column; and

e) eluting the purified Factor VIII polypeptide from the ion exchange column.

- 5 7. A method of claim 6, wherein the desorbing solution of step (b) contains 50% (v/v) ethylene glycol, and the dilution of step (c) is performed using a solution comprising less than 50% (v/v) ethylene glycol, such that the final concentration of ethylene glycol is from about 17% to about 33% (v/v).
8. A method of claim 6, wherein the desorbing solution of step (b) contains 50% (v/v) ethylene glycol, and the dilution of step (c) is performed using a solution comprising no
10 ethylene glycol, such that the final concentration of ethylene glycol is from about 17 to about 33% (v/v).
9. A method of claim 8, wherein the desorbing solution is diluted from about 1.5-fold to about 3-fold.